Processors of agricultural products

ADM

ARCHER DANIELS MIDLAND COMPANY BOX 1470 DECATUR, ILLINOIS 62525 TEL: 217/424-5200

November 22, 2000

BY HAND DELIVERY

Office of Premarket Approval (HFS-200)
Food and Drug Administration
Center for Food Safety and Applied Nutrition
200 C Street, SW
Washington, DC 20204

Re: GRAS Notification- Exemption Claim

Dear Sir or Madam:

Pursuant to proposed 21 C.F.R. § 170.36(c)(1) (62 Fed. Reg. 18938, 18960, April 17, 1997), the Archer Daniels Midland Company ("ADM") hereby claims that plant sterols derived from oil seed processing are Generally Recognized as Safe ("GRAS"); therefore, they are exempt from statutory premarket approval requirements.

This GRAS determination has been made through scientific procedures including a review of the data and information contained in Lipton's GRAS notification submitted as a Food Master File for plant sterol esters and the subsequent Interim Final Rule authorizing a health claim on the association of plant sterol esters and the reduction of the risk of coronary heart disease.

The following information is provided in accordance with the proposed regulation:

Notifier: Archer Daniels Midland Company

Mailing Address: 1001 North Brush College Road
Decatur, IL 62521
Attention: Mark W. Empie, Ph.D.
Vice President Regulatory and Scientific Affairs

GRAS Substance: Plant Sterols Derived from Oil Seed Processing

[redacted]

Intended Use: As a direct ingredient in vegetable oil spreads, dressings for salad, health drinks, health bars, yoghurt-type products, and as a raw material for the manufacture of vegetable oil derived plant sterol esters. The plant sterols are intended to be used for their nutritive value.

Basis for GRAS Determination: Scientific Procedures

A notification package providing a summary of the information which supports this GRAS determination is enclosed with this letter and is being submitted in triplicate. The data and information that are the basis for the ADM GRAS determination are available to the FDA for review and copying upon request.

If you have any questions, please contact me at (217) 424-2463.

Sincerely,

Mark W. Empie, Ph.D.
Vice President Regulatory and Scientific Affairs
Archer Daniels Midland Company

GRAS Notification for

Plant Sterols

Derived from Oil Seed Processing

Provided by: The Archer Daniels Midland Company

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i. Executive Summary

This document is provided in support of Archer Daniels Midland's (ADM) determination that a sterol fraction derived from oil seed processing is Generally Recognized as Safe (GRAS). ADM has developed a purified plant sterol product which may be used as a direct ingredient in vegetable oil spreads (up to 12% use level), health drinks, dressings for salad, yoghurt-type products and health bars at a level of about 1g per serving.

The oil seed derived sterol product may also be used in the manufacture of sterol esters using vegetable oil fatty acids, and these sterol esters may be incorporated in fat based spreads (up to 12% sterol use level), dressings for salad, health drinks, health bars and yoghurt-like products at an equivalent sterol level of about 1 gram sterol per serving.

This GRAS determination is based in part upon the reviews and data contained in a document submitted by Lipton as a Food Master File for plant sterol esters and the subsequent Interim Final Rule authorizing a health claim on the association of plant sterol esters and the reduction of the risk for coronary heart disease. ADM has conducted its own review of the data, as well as literature published since the Lipton information was presented to the Agency. ADM finds that the data supports the GRAS status of plant sterols in the above listed applications.

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I. Introduction

Archer Daniels Midland Company (ADM) submits this document as a notification to FDA of the Generally Recognized as Safe (GRAS) status of oil seed derived plant sterols and sterol esters, pursuant to FDA's policy described in Federal Register Notice of April 17, 1997 (62 FR 18938). Proposed uses of plant sterols are incorporation in vegetable oil spreads (up to 12% use level), dressings for salads, health bars, health drinks and yoghurt-type products. These sterols may also be used as ingredient starting materials for the manufacture of oil seed derived, plant sterol esters to be used in the same application. The anticipated use levels are expected to be 1 gram per serving on a sterol basis.

The basis for this GRAS determination is scientific procedures. Although there has been a past history of use for many years through general consumption of vegetable foods, the proposed uses may increase daily intake of sterols by as much as 5 to 15 times.

During 1985, the Agency accepted a GRAS affirmation file covering vegetable oil sterols which outlined dietary consumption estimates known in the literature at that time. This file was later withdrawn without prejudice by the petitioner, General Mills, Inc. The proposed use requested in this file was as an emulsifier.

On January 11, 1999, Lipton submitted a document to FDA, which was filed as a Food Master File (FMF 625), notifying the Agency of their determination that vegetable oil sterol esters were GRAS for use in vegetable oil spreads. After review of this file, FDA issued a letter to Lipton indicating the Agency had no further questions for Lipton's marketing of the product under a self determination of GRAS (Rulis, 1999).

On February 18, 1999, McNeil submitted a document which was also filed as a Food Master File (FMF 626) which disclosed their determination that plant stanol esters were GRAS for use in vegetable oil spreads and salad dressings.

On February 1, 2000, Lipton submitted to the Agency a proposal to obtain a health claim for the association of plant sterol esters and the reduction in the risk of coronary heart disease. After review, FDA has published an Interim Final Rule permitting a health claim for the relationship between plant sterol esters and a reduction in the risk of coronary heart disease (65 FR 54685). This Interim Rule also included stanol esters, a hydrogenated form of sterol.

On the basis of the totality of the evidence concerning vegetable oil sterols and that for plant sterol esters which are converted in the digestive tract to unesterified sterols, the active component of this petition, ADM concludes that vegetable oil-derived plant sterols are GRAS for use in the listed applications.

II. Background

Oil seed-derived plant sterols consist of a class of compounds composed primarily of β-sitosterol, stigmasterol and campesterol with lesser amounts of brassicasterol, Δ5-avenasterol and several corresponding stanols. In nature, these phytosterols appear as free sterols, esterified forms with fatty acids and phenolic acids, or conjugated with glycosides. Plant sterols are present in many fruits, vegetables, nuts, seeds, cereals and legumes (Weilrauch and Gardner, 1978). Crude food oils from soy, corn, sunflower, canola, palm and other crops may contain

[redacted]

100-500 mg phytosterol per 100 grams of oil. Rice bran, wheat germ and corn fiber oil may also contain measurable amounts.

In the process of refining these crude plant (vegetable) oils, the majority of the sterols are removed into a distillate fraction during the deodorization of the oil. This distillate is the starting point for the recovery in high purity of the subject sterols. This same distillate fraction is also the starting point for the recovery of Vitamin E.

Various studies have been conducted which indicate daily intakes of sterols range from 78 mg/day in the general population to 344 mg/day in lacto-ovo vegetarians (Nair, 1984). The British diet was estimated to contribute 158 mg/day to intake (Morton, 1995), while in Japan estimates of consumption are about 373 mg/day (Hirai, 1986). In Scandinavia, vegetarians consume about 513 mg/day and non-vegetarians 398 mg/day (Mettinen and Tarpila, 1978). Sterol intake among U.S. railroad workers was 170 mg/day (de Vries, 1997). A review by Ling and Jones (1995) estimated the average U.S. intake to be 250 mg/day and higher among vegetarians.

During the 1950's to 1980's, Eli Lilly marketed a drug product made up of primarily β-sitosterol as a treatment for hypercholesterolemia. Numerous high dose clinical studies have been conducted (up to 25,000 mg/day), some over a 3 year period, to assess effects on blood cholesterol levels (Pollak, 1981; Pollak, 1985; Ling, 1995). Over 1800 people participated in these studies including, men, women, adolescents and children. No adverse effects were reported. More recently, plant sterols as the esters have been the subject of additional studies, and there has been general consumption in the U.S. Because plant sterol esters are hydrolyzed in the digestive tract to liberate the active, cholesterol reducing sterols, this data on plant sterol esters is directly applicable for this notification (65 FR 54685 at page 54690; Swell, 1954; Best, 1958; Mattson, 1977).

III. Chemical Identity and Composition

a) Identity

Vegetable oil-derived plant sterols are a mixture of mainly β-sitosterol, stigmasterol and campesterol, obtained from the refining of crude edible vegetable oils. This sterol fraction arises from a distillation of the crude oil with subsequent purification steps to separate the sterols from fatty acids, lecithins and other compounds. Some variation in the relative amounts of each sterol occurs depending on the source of the crude oil and on growing conditions of the oil seeds. However, typical ranges can be found as follows:

[redacted]

Sterol	CAS No.	Range (%)	MW
β-Sitosterol	83-46-5	40-55	414.7
Campesterol	474-62-4	20-28	400.7
Stigmasterol	83-48-7	14-23	412.7
Brassicasterol		0-6	398.6
D5 Avenasterol		0-4	410.6
β-Sitostanol	19466-47-8	0.4	416.7
Campestanol		0-2	
Others	NA	0-5	NA

Structures for each of these compounds and other related phytosterols are found in Appendix A.

b) Properties

Sterols are a yellow semi-solid at room temperature and melt above 60°C. They are insoluble in water and dissolve in oils at about 2 to 3g/100g oil at 37°C. Their solubility in fats increases when the sterol is esterified with fatty acids (Mattson, 1982), and they may be emulsified with surfactants such as lecithin to form liposomes and micelles.

Sterols are minimally absorbed (2-12%) in the digestive tract. That which is absorbed is quickly eliminated and not metabolized. Consequently, sterols are not a source of energy.

IV. Production Methods and Products

Phytosterols are a byproduct of plant and vegetable oil refining. Oil seeds, such as corn, palm, soy, rape and sunflower, are pressed or extracted to obtain the oil. This oil undergoes a series of refining processes to remove lecithins, free fatty acids, color bodies, off-odors and off-flavors. One step deodorizes the oil by steam distillation at reduced pressure, and the resulting distillate contains the phytosterol fraction. This deodorizer distillate is also the raw material used in the manufacture of U.S.P. grade, natural-source Vitamin E. Oil seed derived sterols and Vitamin E represent about 0.3 to 0.4% of the oil volume, while the deodorizer distillate contains up to 30% tocopherols and sterols. The use of these steps in vegetable oil purification have been in common use for many years, and additional information about oil refining and resulting co-products may be found in Hui (1996).

a) Oilseed Sterol Purification

In a series of steps Vitamin E tocopherols are separated from the sterols. Typically the deodorized distillate is subjected to a further distillative stripping of free fatty acids. The undistilled material is subjected to an ethanolysis transesterification step to form fatty acid ethylesters and glycerol from the triglycerides, and free phytosterols and fatty acid ethylesters from the sterol esters. Glycerol is removed, and the fatty acid esters are stripped out. Phytosterols and tocopherols are then distilled from the remaining mixture. The phytosterols are

[redacted]

separated from the tocopherols by crystallization from a heptane solution, and further purified through recrystallization. The final product may be packaged in bulk, prilled or micronized to a fine powder. These products are also suitable for use in the manufacture of sterol esters or for conversion to other materials by the pharmaceutical industry. The purification outlined above for Vitamin E and sterols has been in use for many years and practiced by ADM for the last 4 years. ADM is the world's largest producer of natural-source Vitamin E.

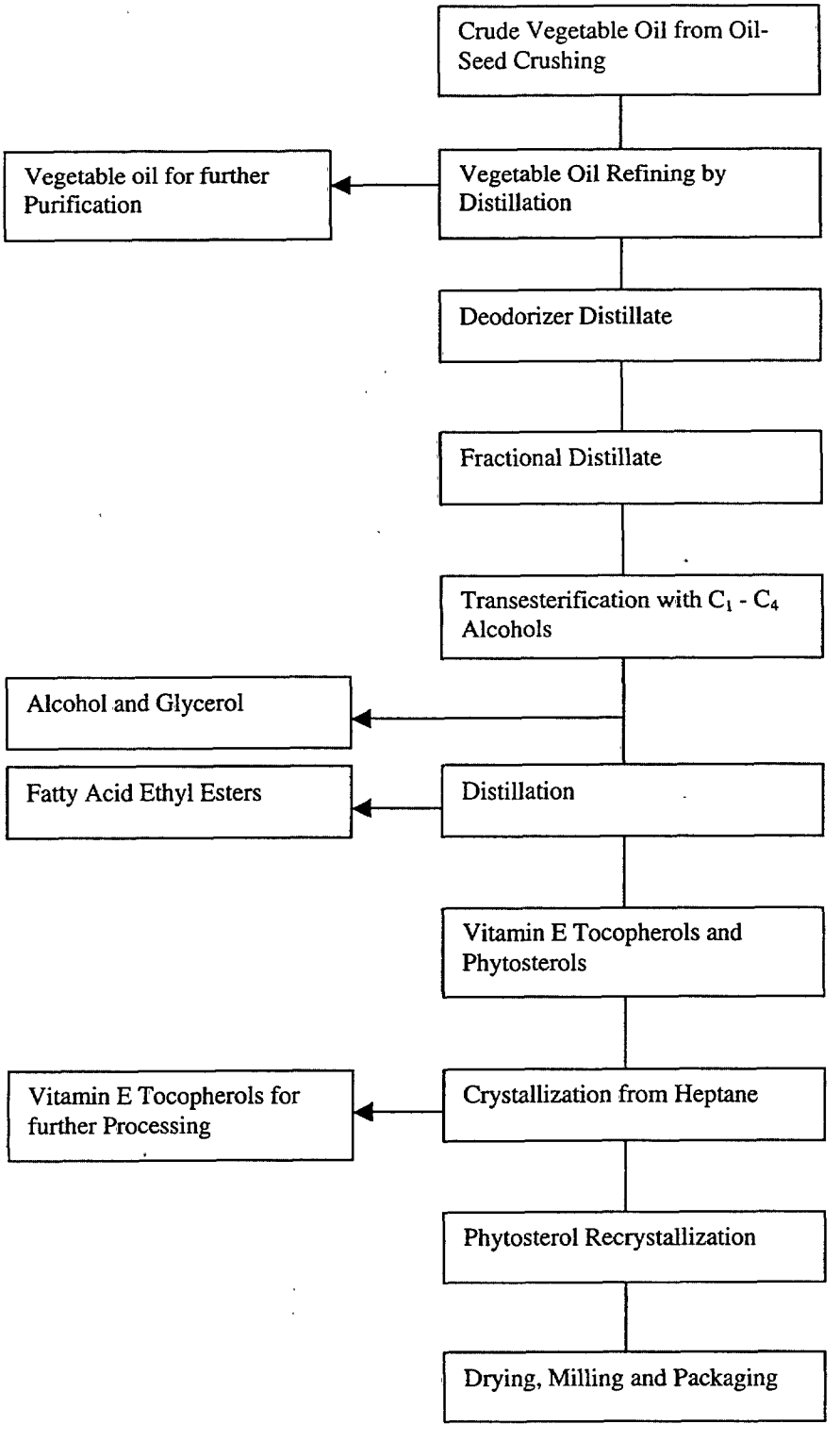
Oil seed-derived sterol esters will be manufactured from food grade vegetable oil fatty acids and triglycerides using suitable food grade procedures and methods for esterification or transesterification.

A process flow diagram is given in figure 2.

[redacted]

Figure 2

Process Diagram for Phytosterol Manufacture



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b) Quality Assurance

All materials are produced according to current food-grade Good Manufacturing Practice (cGMP; 21 CFR 110 and 111).

c) Proposed Food-grade Specification

The oil seed-derived sterols obtained by the above described process will conform to the following specifications. The ranges found are caused by the natural variation in phytosterol content arising from the different input oilseed crops and from year-to-year growing condition variations. The specification provided represents the typical range for production over the course of one year. ADM phytosterols are Kosher and Pareve.

Phytosterol	Range (weight %)	Analysis Method
Total Phytosterols	Minimum 90%	ADM E-0110
β-Sitosterol	40-58%	See Appendix B
Campesterol	20-30%	
Stigmasterol	14-22%	
Brassicasterol	0-6%	
Sitostanol	0-5%	
Appearance:	Granular Solid	
Odor:	Slight	
Tocopherols:	0-15 mg/g	
Melting Point:	135-145C	
Heavy Metals:	Maximum 10ppm	
Lead	<1ppm	
Solubility in Water:	Insoluble	

d) Representative Production Batches

ADM has produced vegetable oil sterols commercially for over two years. The following analyses are representative of products arising from this production.

	Batch 1	Batch 2	Batch 3	Batch 4	Batch 5
Production Date:	11/18/99	1/18/00	3/4/00	8/1/00	10/8/00
Total Phytosterol:	98.7	95.4	96.2	98.1	95.4
Sitosterol	48.2	46.5	47.2	47.1	46.7
Campesterol	27.2	26.2	26.6	26.3	26.6
Stigmasterol	16.1	16.6	15.0	18.3	15.9
Brassicasterol	4.9	3.8	5.0	4.1	3.9
Sitostanol	2.3	2.3	2.4	2.3	2.3
Total votatiles:	<2.0	<2.0	<2.0	<2.0	<2.0
Ash:	<0.1	<0.1	<0.1	<0.1	<0.1
Heavy Metals:	<10 ppm	<10 ppm	<10 ppm	<10 ppm	<10 ppm

e) Complete Product Characterization

The phytosterol specification establishes the composition to be a minimum 90% by weight. A number of other minor oil constituents arise which remain after the purification process. These include mono-, di and triglycerides, wax esters and squalene. In addition, batches have been tested for pesticide residues which might be found on the oilseed crops. No residues have been found for 20 organochlorine compounds analyzed (Appendix C).

V. Intended Use in Food

Although the subject plant oil sterols are commonly found in food, the purpose of this GRAS notification is to advise the Agency of the intended addition of oil seed derived sterols or their corresponding fatty acid esters to certain foods. Specifically, the intended use is proposed for incorporation into fat based spreads, dressings for salad, health bars, health drinks, and yoghurt-like products. Additionally, ADM sterols will be used as a food grade ingredient in the manufacture of vegetable oil derived plant sterol esters for use in the proposed GRAS uses.

Food ingredients provide nutritive value, flavor, odor or technical effects in food. Although a previous GRAS submission proposed a technical use as an emulsifier, the present notification is for nutritive value. FDA has noted that nutritive value encompasses assisting in the efficient functioning of classical nutritional processes and of other metabolic processes necessary for the normal maintenance of human existence (65 FR 54688).

The Agency has issued an interim final rule permitting the use of a health claim relating the consumption of plant sterol esters with a reduction in the risk of coronary heart disease. The mechanism associated with this claim is the lowering of serum cholesterol. The Agency has evaluated 15 studies conducted between 1953 and 2000 to elucidate the relationship of serum cholesterol and consumption of plant sterols and plant sterol esters. In evaluation of the data

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from both free and ester forms, the Agency acknowledged that plant sterol esters are hydrolyzed in the gastrointestinal tract to free sterols, and agreed that the free sterol is the active moiety in reducing serum cholesterol (65 FR 54690). Consequently, the nutritive value definition for a food ingredient is met. The notified sterol substance is the active moiety in reducing serum cholesterol, and this helps to maintain normal digestive and cardiovascular processes.

VI. Consumer Exposure

ADM intends to provide to customers plant sterols and sterol esters for incorporation into spreads, dressings for salad, yoghurt-type products, health drinks and health bars. The main sources for plant sterols in the human diet arise from cooking oil and margarines (0.3-0.5%) (Morton, 1995), breads and cereals, vegetables and fruit (<0.05% to 0.2%). Some seeds such as sunflower and sesame contain 0.5-0.7%, while legumes can contain 0.22% phytosterols (Lange, 1950, Weihrauch, 1978). As mentioned, the typical U.S. diet provides about 250 milligrams of phytosterols per day (Nair, 1984; Ling, 1995).

a) Spreads

The oil seed derived sterol products are expected to compete directly with plant sterol and stanol esters for this use category. Consequently, exposure calculations made for these products are directly applicable for free-sterols after correcting for the fatty acid side chain. Daily consumption of vegetable oil spreads is between 9 grams and 28 grams, with typical intake around 12 to 14 grams per day (USDA 1997). Incorporation of up to 12% free sterols in a reduced-fat spread would lead to an intake of sterols of about 1.1 to 3.4 grams per person per day (0.12 x 28g spread for the 90th percentile). This represents a 4 to 14-fold increase over the 250 mg/day intake from food sources.

b) Dressings for Salad

The inclusion of the sterol products in dressings for salad is as a replacement for currently marketed sterol and stanol esters in this application. Consequently, this does not represent an additional category. Lipton has estimated that adult consumption of plant sterols from reduced fat dressings is in the range of 1.1 to 2.2 grams per day. Consumption by children would be lower. At the 90th percentile level, consumption by adults of both spreads and dressings would contribute 4.4 grams sterols per day in the diet. Children would be lower.

c) Yoghurt-Type Products

Use in yoghurt-type products is expected to be 1 gram per day. This is based on the expected incorporation of 1 gram plant sterols per yoghurt container and consumption of 1

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container per day. Yoghurt-type products may include dairy-based, soy-based and other protein-based formulations.

d) Health Bars

Inclusion of oil seed derived sterol products in health bars may be achieved at a 1 gram sterol equivalent per bar level. The food matrix itself does not limit the inclusion rate per se, however, higher levels will impart a chalky mouth-feel which will be unacceptable for the consumer. Serving size for health bars is 40 grams, and the daily consumption is between 1 to 3% of all users. At a 1 gram per bar level, all user, all age group, mean consumption of sterols would be 0.6 grams. For the 90th percentile, consumption of sterols would be 1.0 grams.

e) Health Drinks

Sterol product inclusion in health drinks is proposed at a 1 gram sterol equivalent per serving level. These products include liquid and reconstituted powder meal replacements and dry powder non-reconstituted products, as well as fortified sport drinks. Serving size is 240 mls. The food matrix may limit the inclusion rate of sterols due to the suspendability and the mouth feel of the product. The percentage of users is between 0.87 and 3.7% for all age groups (>12 years). At a 1 gram per serving level, all user, all age group consumption of sterols would be 1.5 grams for the mean and 2.8 grams for the 90th percentile.

f) Consumption by Other Population Groups

Generally, consumption of plant sterols by populations in the U.S. and in Europe range from 250 mg per day to 300 mg per day. Intakes by adult vegetarians and their children are slightly higher (340 to 513 mg/day; Nair, 1984; Miettiner, 1978). Infants fed formula can consume 2- to 4-fold more phytosterols than adults on a body weight basis (Huisman, 1996). The proposed uses in this notification are not directed toward infants. Lastly, individuals with hypercholesterolemia have been treated with plant sterols. Numerous clinical trials in this population sub-group have been run without reports of adverse effects. Studies have lasted from several months to up to 4 years with intakes of up to 25 grams per day (reviewed by Pollak and Kritchevsky, 1981; Pollak, 1985; Ling, 1995). Ingestion of moderate levels of plant sterols are reviewed as safe to lower blood cholesterol.

VII. Safety Information

On January 11, 1999, Lipton notified FDA of its determination that plant (vegetable oil) sterol esters are GRAS (Franke, 1999, p 000030). This notification presented a thorough evaluation of the available literature, several unpublished studies which have subsequently been published (Baker, 1999; Hepburn, 1999; Waallkens-Berendsen, 1999; Weststrate, 1999; Ayesh, 1999; Hendricks, 1999; Sanders, 2000) and information from an outside panel of experts retained

by Lipton to evaluate the safety data (Franke, 1999, p 000057). This information drew upon studies which included both sterol esters and free sterols. FDA in the Interim Rule establishing a health claim for CHD and sterol and stanol esters agreed with Lipton that the active moiety for serum cholesterol reduction is the free sterol (65 FR 54685 at page 54690). Lipton also asserted that sterol esters are converted in the gastrointestinal tract to free sterols. ADM agrees with both conclusions. Consequently, as the data on free sterols supports the GRAS determination for sterol esters, the sterol ester literature may be used to support the safety and ADM's GRAS determination for free-sterols. References and summaries contained in Lipton's notice provide a compilation of relevant information on safety and use (Franke, 1999, pp 000030-000061). ADM is in agreement with the safety summaries and conclusions in the Lipton notification.

a) **Reviews Pertaining to Safe Use**

Concise summaries of Lipton's most relevant safety information are provided in: 1) an April 30, 1999 memo from FDA Consumer Safety Officer, Dr. Paulette M. Gaynor to the Administrative File FMF 000625, 2) an April 30, 1999 internal FDA memo from Dr. Linda Pellicore to Dr. Linda Kahl, 3) an April 9, 1999 letter from Lipton's Dr. William Franke to Dr. George Pauli, 4) and as a report from the Scientific Committee for Foodstuffs regarding Lipton's submission for sterol ester margarine under the European Union's Novel Foods Regulation (258/97/EC).

These documents summarize:

1. Absorption, distribution, metabolism and excretion (ADME) (Gaynor, pg 4, Pellicore, pg 1, Scientific Committee, pg 7).
2. Clinical studies, including those related to the drug approval of Cytellin (Gaynor, pg 5, Pellicore, pg 1, Scientific Committee, pg 6).
3. Effects on fat soluble vitamins and other concerns (Gaynor, pg 6, Pellicore, pg 2, Franke, 1999a, pg 1, Scientific Committee, pg 8).
4. Reproduction toxicity and estrogenic effects (Gaynor, pg 7, Pellicore, pg 4, Scientific Committee, pg 10).
5. Subchronic animal toxicity, genotoxicity and carcinogenicity (Gaynor, pg 9, Pellicore, pg 4, Scientific Committee, pg 9).

Lipton's GRAS Panel consisted of Drs. Borzelleca, Glinsmann, Kritchevsky and Pariza (Franke, 1999, p 000057). The Panel concluded that the extensive human safety information on phytosterols arising from the free sterol literature was pertinent to establishing safe exposure limits for sterol esters. The numerous (free sterol) published clinical studies including high-dose (up to 25,000 mg/day), long term (up to 3 years) exposures demonstrated safety (Ling, 1995; Pollak, 1981, Pollak, 1985). Over 1800 people participated in these trials and some studies included adolescents and children. No adverse health effects were reported.

The Panel concluded that an acceptable ADI was established as 130 mg/kg/day (as the free phytosterol, Franke, 1999, p 000058). Consumption of a combination of products in the proposed categories of ADM's GRAS notification are within the range of this ADI.

The members of the European Union's Scientific Committee for Foodstuffs similarly concluded that: 1) based on extensive toxicological testing of phytosterol preparations in a 13-

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week rat feeding study, in a two-generation rat feeding study, in studies of estrogenic potential and in tests of genotoxicity, no safety concerns were apparent. The phytosterol profile judged to be safe encompasses the specification of the ADM product. The Committee also indicated that the use level of phytosterol esters in margarine should be set at 8% (as the free sterol). The consumption level of margarine in Europe is between 20 to 30 grams per day, which leads to a consumption of 1.6-2.4 grams of sterols per day in this application.

b) Daily Consumption Estimate

The total daily consumption may be calculated for a group consuming all the listed products. Using the mean and 90th percentile values, this calculation yields a value of 5.5 for the mean and 10.6 for the 90th percentile.

	Amount (grams)	
	Mean	90th %
From conventional food	0.25	(0.25)
Spreads	1.1	3.4
Dressings for salad	1.1	2.2
Yoghurt-type products (estimate)	1.0	(1.0)
Health bars	0.6	1.0
Health drinks	1.5	2.8
	5.5	10.6

Based on the 130 mg/d ADI, an acceptable consumption level for a 70 kg person would be 9.1 grams. The mean group consuming all products is well within the ADI value at 5.5 g/day. At the 90th percentile, the consumption calculated is 16% higher than the ADI calculated by Lipton. As a review of this ADI determination, the value is based on a 90-day rodent feeding study of sterol esters where the doses fed to male rats were 3900 mg/kg and to female rats were 4200 mg/kg. The male dosage was taken for the NOAEL and a safety factor was set as a logarithm value halfway (=30) between the 10 times and 100 times comparison to the NOAEL.

Additional literature studies may be cited which show this NOAEL value, and hence, the ADI is a conservative number. Between 1954 and 1982, Eli Lilly Research Laboratories marketed phytosterols in the U.S. and Canada. The predominant sterol present was free sitosterol. In 1970, the National Formulary (1970) published a monograph on sitosterol. The dosages recommended were 3 grams taken 3 times a day with the dosage range being from 9 grams to 30 grams per day. In 1973, the U.S. Dispensatory (Osol, 1973) published a monograph stating the usual dose for sitosterols was 9 g/day and that the dose may be expanded to 30 grams or more. No concerning adverse events have been reported for these use levels.

Several toxicological studies have been conducted on the Lilly product which include 5% additions to diets fed to rats for 18 months. Similarly, dogs were fed 1g/kg-bw for 18 months. In both cases the animals gained weight and had no visceral damage. Similar dose level feeding studies conducted by Shipley (1958) in rats, dogs and rabbits for about 2 years in duration showed no alterations in growth or pathologic changes due to test material. The sum total of the data consistently show no adverse effects in any animal or human species at high dose level.

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The safety factor for the 90th percentile for a person consuming all ADM proposed products is 26, as compared to 30 in the Lipton ADI calculation. For the 50th percentile a safety factor of 50 is calculated. The consumption of the usual dose of 9g/day and higher doses for the Cytellin product and the lack of adverse effects in numerous long term, high dose animal studies show that the Lipton ADI may be conservative and exceeding this value by 16% is justified. Consequently, we calculate that the proposed uses and use levels are within the safety requirements for GRAS use.

VIII. Conclusion

Based upon the information summarized in this notification, Archer Daniels Midland determines that plant sterols and sterol esters derived from oilseed refining for use in 1) vegetable oil spreads, 2) dressings for salad, 3) health bars, 4) yoghurt-type products, 5) health drinks and 6) vegetable oil sterol ester manufacture are generally recognized as safe within the meaning of §201 of the Federal Food, Drug and Cosmetic Act; 21 CFR §§170.3 and 170.30; and the proposed rule listed at 62 Fed. Reg. 18960.

IX. References*

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2. Baker, V.A., Hepburn, P.A., Kennedy, S.J., Jones, P.A., Lea, L.J., Sumpter, J.P. and Ashby, J. (1999). "Safety evaluation of phytosterol esters. Part 1. Assessment of oestrogenicity using a combination of In vivo and In vitro assays." Food and Chemical Toxicology, 37, pp 13-22.

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4. deVries, J.H.M., Jansen, A., Kromhout, D., van de Bovenkamp, P., van Stavern, W.A., Mensink, R.P. and Katan, M.D (1997). "The fatty acid and sterol content of food composites of middle-aged men in seven countries," Journal of Food Composition and Analysis, vol. 10, pp 115-141.

5. 62 Federal Register 18938, (April 17, 1997). "Substances generally recognized as safe; proposed rule".

6. 65 Federal Register 54685 (September 8, 2000). "Food labeling: health claims; plant sterol/stanol esters and coronary heart disease; interim final rule".

7. Franke, W., (January 11, 1999). Submission to FDA Office of Premarket Approval, "GRAS notification for vegetable oil sterol esters used in vegetable oil spreads".

8. Franke, W., (April 9, 1999a), FDA, Letter to Dr. George Pauli.

9. Gaynor, P.M., (April 30, 1999), FDA, Memo to Administrative File FMF 000625.

10. Hendricks, H.F.J., Westrate, J.A., van Vliet, T. and Meijer, G.W. (1999). "Spreads enriched with three different levels of vegetable oil sterols and the degree of cholesterol lowering in normocholesterolaemic and mildly hypercholesterolaemic subjects". European Journal of Clinical Nutrition, 53, pp 319-327.

11. Hepburn, P.A., Horner, S.A. and Smith, M. (1999). "Safety evaluation of phytosterol esters. Part 2. Subchronic 90-day oral toxicity study on phytosterol esters - A novel functional food." Food and Chemical Toxicology, 37, pp 521-532.

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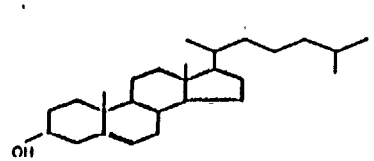
* For additional references, see bibliographies at the end of the Lipton submission.

[redacted]

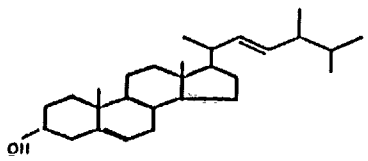
Appendix A

Structures of Plant Sterols

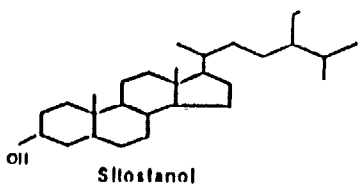
[redacted]



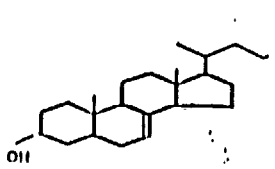
Cholesterol



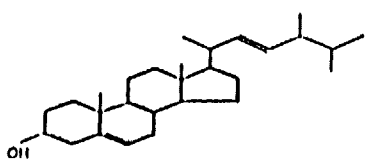
Stigmasterol



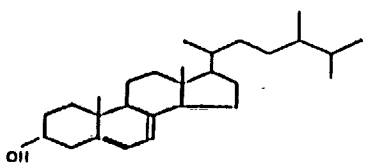
Sitostanol



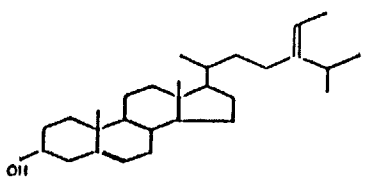
D7-Avenasterol



Brassicasterol



D7-Campesterol



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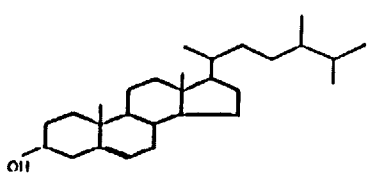
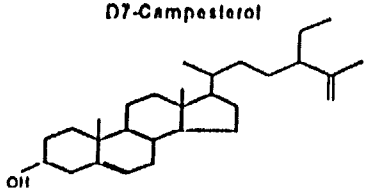
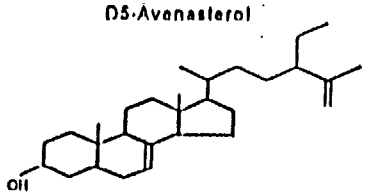


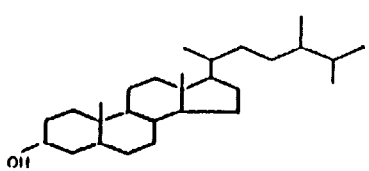
Campesterol



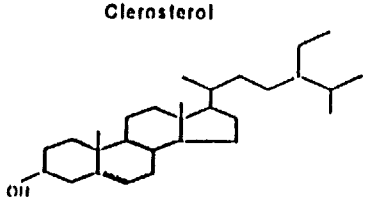
Clerosterol



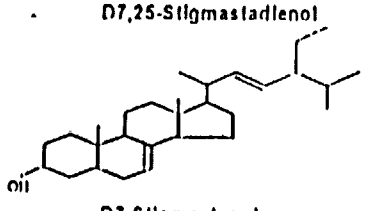
D7,25-Stigmastadienol



Campestanol



β-Sitosterol



D7-Stigmastenol

Appendix B

ADM Sterol Analysis Procedure

[redacted]

ADM FOOD ADDITIVES VITAMIN E	ANALYTICAL PROCEDURE DETERMINATION OF FREE STEROLS BY GAS CHROMATOGRAPHY	1.0	E-0110
		DATE ISSUED 12-01-95	PAGE 1 OF 5
		SUPERSEDES NONE	

1.0 SCOPE

THIS PROCEDURE DEFINES THE METHOD FOR THE DETERMINATION OF FREE STEROLS BY CAPILLARY GAS CHROMATOGRAPHY. SAMPLES ARE SILYLATED AND CHROMATOGRAPHED AS TRIMETHYSILYL ESTERS. TOCOPHEROLS MAY BE DETERMINED SIMULTANEOUSLY PER E-0101.

2.0 APPLICABLE DOCUMENTS

2.1 DPI-A-MI-G-GC-98-2 DETERMINATION OF ... STEROLS
2.2 E-0101 DETERMINATION OF TOCOPHEROLS BY GAS CHROMATOGRAPHY
2.3 E-0116 SAPONIFICATION OF DEODORIZER DISTILLATE

3.0 SAFETY

3.1 PROPER VENTILATION SHOULD BE USED AT ALL TIMES WHEN WORKING WITH THE CHEMICALS REQUIRED FOR THIS PROCEDURE.

4.0 REAGENTS

4.1 PYRIDINE, LOW MOISTURE
4.2 BSTFA + TMCS 99:1 SILYLATING REAGENT
4.3 HEPTADECANYL STEARATE INTERNAL STANDARD SOLUTION (HDS) - HEPTADECANYL STEARATE, ACCURATELY WEIGHED, IN TOLUENE FOR FINAL CONCENTRATION OF 1.00 +/- 0.02 mg/ml.
4.4 STIGMASTEROL STANDARD OF KNOWN PURITY

5.0 EQUIPMENT AND APPARATUS

5.1 GAS CHROMATOGRAPH WITH AUTOSAMPLER AND FID DETECTOR, HELIUM CARRIER GAS, SUPELCO SPB-5 (30 m, 0.25 mm ID, 0.25 um film thickness) COLUMN OR EQUIVALENT.
5.2 ANALYTICAL BALANCE ACCURATE TO 0.1 mg.
5.3 VIAL, SCREW CAP, 10 ml CAPACITY MINIMUM
5.4 DISPENSERS OR PIPETS, 1 ml AND 5 ml.
5.5 DRY HEATER APPROPRIATE FOR VIALS IN 5.3.
5.6 AUTOSAMPLER VIALS

6.0 PROCEDURE

000028

6.1 SOURCES OF ERROR IN THIS PROCEDURE INCLUDE: INACCURATE WEIGHTS, INCOMPLETE DERIVATIZATION, CONTAMINATION BY WATER, INTERFERENCE IN STEROL OR INTERNAL STANDARD PEAKS, DEGRADATION OF STANDARDS, AND EQUIPMENT PROBLEMS. CONSULT THE LAB SUPERVISOR AS REQUIRED.

[redacted]

APPROVAL	LABORATORY 12-1-95	DIRECTOR QUALITY 12/1/95	PRODUCTION 12/1/95	EFFECTIVE DATE 12-01-95

ADM

FOOD ADDITIVES

VITAMIN E

ANALYTICAL PROCEDURE

DETERMINATION OF FREE STEROLS BY GAS CHROMATOGRAPHY

1.0	E-0110
DATE ISSUED 12-01-95	PAGE 2 OF 5
SUPERSEDES NONE	

6.2 DETERMINATION OF STEROL RESPONSE FACTOR

6.2.2 AS A MINIMUM, THE STEROL RESPONSE FACTOR IS DETERMINED AFTER EACH HELIUM CHANGE, FOR EACH NEW BATCH OF INTERNAL STANDARD SOLUTION, AND AFTER ANY EQUIPMENT MAINTENANCE.

6.2.2 THE STIGMASTEROL STANDARD WEIGHT IS 10 TO 15 mg. RUN AT LEAST TWO STANDARDS. PROCEED TO PARAGRAPH 6.4 FOR ANALYTICAL PROCEDURE.

6.3 DETERMINE SAMPLE WEIGHT PER THE FOLLOWING GUIDELINES:

DEODORIZER DISTILLATE, BEFORE AND AFTER SAPONIFICATION	180-200 mg
STEROL FILTRATION SAMPLES	40-50 mg
STEROLS, PRILLED	40-50 mg

(THE WEIGHT FOR MOST OTHER PROCESS SAMPLES WILL BE DETERMINED BASED ON THE TOCOPHEROL CONTENT.)

6.4 WEIGH STANDARD OR SAMPLE INTO A SCREW CAP VIAL. RECORD THE WEIGHT TO THE NEAREST 0.1 mg. (A 5 PLACE BALANCE IS AVAILABLE FOR THE STANDARDS.)

6.5 BY MEANS OF A PIPET OR DISPENSER, ADD 1 ml PYRIDINE AND 1 ml OF SILYLATING REAGENT. CAP AND SHAKE WELL.

6.6 SET A TIMER AND HEAT IN A DRY BLOCK HEATER SET AT 80C FOR 20 MINUTES. SHAKE AS NECESSARY.

6.7 REMOVE FROM HEAT AND BY MEANS OF A PIPET OR DISPENSER, ADD 5 mls OF HDS INTERNAL STANDARD SOLUTION. SHAKE WELL.

6.8 TRANSFER TO AN AUTOSAMPLER VIAL AND CRIMP.

6.9 PERFORM GAS CHROMATOGRAPHY PER THE FOLLOWING CONDITIONS:

CARRIER GAS	HELIUM
CARRIER GAS PRESSURE	15 psig
DETECTOR	FID
COLUMN	SUPELCO SPB-5 (30 m, 0.25 mm ID, 0.25 um film) OR EQUIVALENT
INJECTOR TEMPERATURE	280C
DETECTOR TEMPERATURE	330C
SPLIT	110
TEMPERATURE PROGRAM	280C FOR 0 MIN RAMP 10C/MIN TO 300C, HOLD 5 MIN RAMP 1C/MIN TO 310C, HOLD 7 MIN

6.10 CALCULATION

6.10.1 REFER TO FIGURE 1 TO IDENTIFY PEAKS.

6.10.2 CALCULATE STEROL RESPONSE FACTOR AS FOLLOWS:

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[redacted]

$$RF = \frac{(Ais)\ (Ws)\ (Stigmasterol\ purity^*)}{(As)\ (Wis)}$$

WHERE
- Ais = PEAK AREA OF INTERNAL STANDARD
- Ws = WEIGHT OF STIGMASTEROL STANDARD, mg
- *purity = PURITY OF STIGMASTEROL STANDARD IN DECIMAL FORM
- As = PEAK AREA OF STIGMASTEROL STANDARD
- Wis = WEIGHT OF HDS IN 5 ml OF INTERNAL STANDARD SOLUTION, mg

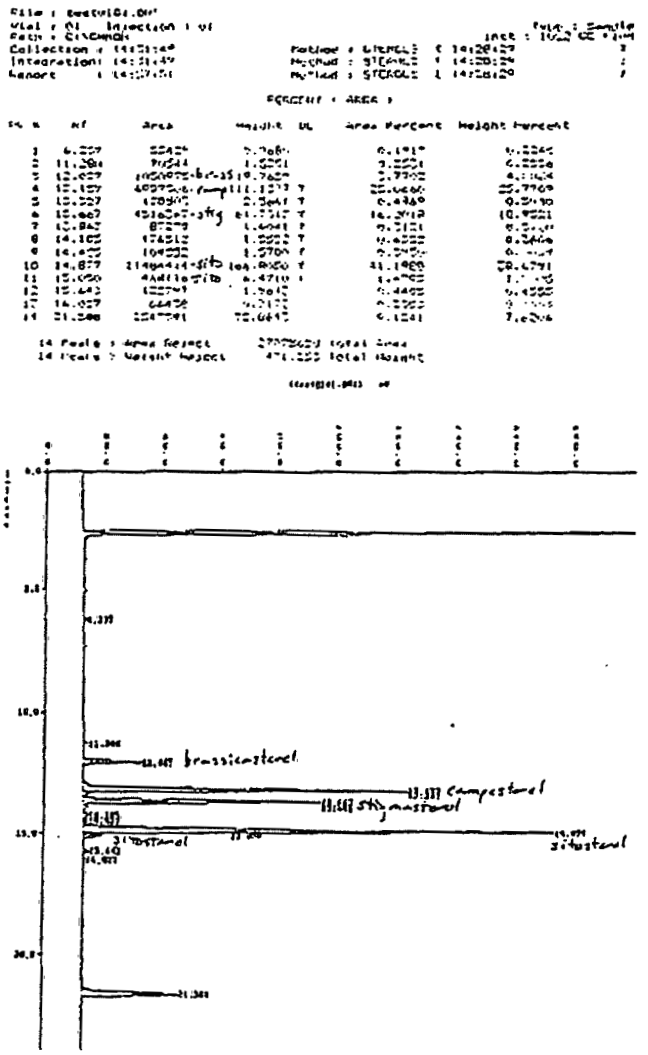
6.10.3 DETERMINE THE STEROL CONTENT IN THE SAMPLE AS FOLLOWS:

$$STEROLS,\ \% = \frac{(As)\ (RF)\ (Wis)\ (100)}{(Ais)\ (Ws)}$$

WHERE
- As = PEAK AREA OF DESIRED STEROL
- RF = STEROL RESPONSE FACTOR FROM 6.10.2
- Wis = WEIGHT OF HDS IN 5 mls OF INTERNAL STANDARD SOLUTION, mg
- Ais = PEAK AREA OF INTERNAL STANDARD
- Ws = WEIGHT OF SAMPLE, mg

ADM FOOD ADDITIVES VITAMIN E	ANALYTICAL PROCEDURE DETERMINATION OF FREE STEROLS BY GAS CHROMATOGRAPHY	1.0	E-0110
		DATE ISSUED 12-01-95	PAGE 4 OF 5
		SUPERSEDES NONE	

FIGURE 1



ANALYTICAL PROCEDURE

ADM

FOOD ADDITIVES
VITAMIN E

DETERMINATION OF FREE STEROLS BY GAS CHROMATOGRAPHY

1.0	E-0110
DATE ISSUED 12-01-95	PAGE 5 OF 5
SUPERSEDES NONE	

VERSION	DATE	DESCRIPTION
1.0	12/01/95	Original Issue

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Appendix C

Pesticide Residue Analysis

[redacted]

Suburban Laboratories, Inc.
4140 Litt Drive Hillside, IL 60162 (708) 544-3260

Date: *Wednesday, August 16, 20*

CLIENT: ADM-Vitamin E Plant
Project: Pesticide Analysis

Lab Order #: *0008287*

Lab Sample ID: 0008287-01 **Date Received:** 08/11/2000 **Collection Date:** 08/08/2000
Client Sample ID: PA0800-001 **Matrix:** PRODUCT

Analyses -PS0200-40	Result	Qual	MDL	PQL	Units	DF	Date Analyzed
ORGANOCHLORINE PESTICIDES		Method:	SW8081A				Analyst: JB
4,4-DDE	ND		40.0	120	µg/Kg	1	08/16/2000
4,4-DDT	ND		20.0	60.0	µg/Kg	1	08/15/2000
Aldrin	ND		40.0	120	µg/Kg	1	08/15/2000
alpha-BHC	ND		40.0	120	µg/Kg	1	08/15/2000
beta-BHC	ND		60.0	180	µg/Kg	1	08/15/2000
Chlordane	ND		1,000	3,000	µg/Kg	1	08/15/2000
delta-BHC	ND		20.0	60.0	µg/Kg	1	08/15/2000
Dieldrin	ND		120	360	µg/Kg	1	08/15/2000
Endosulfan I	ND		40.0	120	µg/Kg	1	08/15/2000
Endosulfan II	ND		30.0	90.0	µg/Kg	1	08/15/2000
Endosulfan sulfate	ND		40.0	120	µg/Kg	1	08/15/2000
Endrin	ND		40.0	120	µg/Kg	1	08/15/2000
Endrin aldehyde	ND		220	660	µg/Kg	1	08/15/2000
Endrin ketone	ND		20.0	60.0	µg/Kg	1	08/15/2000
gamma-BHC	ND		20.0	60.0	µg/Kg	1	08/15/2000
gamma-Chlordane	ND		40.0	120	µg/Kg	1	08/15/2000
Heptachlor	ND		10.0	30.0	µg/Kg	1	08/15/2000
Heptachlor epoxide	ND		60.0	180	µg/Kg	1	08/15/2000
Methoxychlor	ND		260	780	µg/Kg	1	08/15/2000
Toxaphene	ND		2,000	6,000	µg/Kg	1	08/15/2000
Surr: Tetrachloro-m-xylene	101		0	31-146	%REC	1	08/15/2000

Qualifiers:
ND - Not Detected at the Method Detection Limit
J - Estimated or analyte detected below quantitation limit
B - Analyte detected in the associated Method Blank
* - Value exceeds Maximum Contaminant Level
S - Spike Recovery outside accepted recovery limits
R - RPD outside accepted recovery limits
E - Value above quantitation range
H - Analysis run past method holding time

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[redacted]

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SUBMISSION END

Processors of agricultural products

ADM

AM

ARCHER DANIELS MIDLAND COMPANY BOX 1470 DECATUR, ILLINOIS 62525 TEL: 217/424-5200

February 28, 2001

Dr. Paulette Gaynor (HFS-215)
Office of Premarket Approval
Center for Food Safety & Applied Nutrition
Food and Drug Administration
200 C Street S.W.
Washington, D.C. 20204

RE: GRAS Notice (GRN) No. 000061

Dear Dr. Gaynor:

Pursuant to our telephone call of February 2, 2001, Archer Daniels Midland Company (ADM) is providing the following information to supplement our GRAS Notification on vegetable oil-derived plant sterols, submitted on November 22, 2000. During this telephone conversation, the Agency requested clarification on specific use levels in the stated applications, a statement of source for the fatty acids used in sterol ester manufacture, final product specifications for sterol esters, and a statement on how use of the product will be monitored. Our responses are as follows:

Use Levels

Food	sterol use level (per serving)	sterol ester use level
Vegetable Oil Spread	1 g per tablespoon	1.65 g
Dressings for Salad	1 g per serving	1.65 g
Yoghurt-Type Products	1 g per container	1.65 g
Health Bar	1 g per bar	1.65 g
Health Drink	1 g per 8 oz.	1.65 g

Fatty Acid Source

The sterol esters will be produced under food GMP conditions using procedures common in the fats and oils industry for transesterification and direct esterification reactions. The fatty acids will be derived from ADM's oil seed crop, vegetable oil processing activities and may include sources from corn, soybean, sunflower, peanut, cottonseed, safflower, canola and palm sources. The preferred sources are from soybean, sunflower, safflower and canola.

Specification for Sterol Ester Product

Total Sterol Esters*	minimum 90% of total sterols
β Sitosterol	40-58% of total sterols
Campesterol	20-30% of total sterols
Stigmasterol	14-22% of total sterols
Brassicasterol	0-6% of total sterols
Sitostanol	0-5% of total sterols

*Sterol esters will be composed of approximately 60% sterols with fatty acid ester component being about 40%.

Free Fatty Acids	<5%
Odor	slight
Heavy Metals (total)	<10 ppm
Lead	<0.1 ppm
Arsenic	<0.1 ppm
Mercury	<0.1 ppm
Cadmium	<0.1 ppm

The lead specification of less than 0.1 will be applied to the ADM free sterol product as well.

Monitoring

ADM produces the sterol and sterol ester products as a bulk ingredient to our customers, who then formulate and sell a consumer product to the general public. ADM tabulates volume data on sales to our customers, but we do not have access to our customers' sales data to the retail sector. From time to time, we may purchase market scan reports on retail consumer purchases. Upon request by the agency, ADM is willing to share our production and sales data for the purpose of monitoring usage of our sterol products. However, a more detailed analysis of retail product consumption is best obtained from other sources.

We hope the foregoing provides the Agency with the requested information. If you have any further questions, please feel free to contact me.

Very truly yours,

Mark W. Empie, Ph.D.
Vice President,
Regulatory & Scientific Affairs

MWE/jm